JARQ 58 (4), 193-196 (2024) https://www.jircas.go.jp

REVIEW

Depot-Specific Differences in Adipogenesis in Wagyu Cattle

Tomoya YAMADA[1]*, Mitsuru KAMIYA[2] and Mikito HIGUCHI[2]

[1] Western Region Agricultural Research Center, National Agriculture and Food Research Organization, Oda, Japan
[2] Institute of Livestock and Grassland Science, National Agriculture and Food Research Organization, Nasushiobara, Japan

Abstract

Fat depot-specific differences in adipose tissue growth (adipogenesis) are an important factor in the production of beef cattle. Japanese black (Wagyu) cattle are characterized by high adipogenic ability. This review will discuss fat depot-specific differences in adipogenesis among subcutaneous, visceral, and intramuscular depots of fattening Wagyu based on our previous studies. We show that anatomical sites of adipose tissue affect adipocyte size. We also show that fat depot-specific differences in adipogenesis are affected by the expression of adipogenic transcription factors. In addition, we show that fat depot-specific differences in adipokine gene expression are affected by adipocyte size. Furthermore, we show that macrophages infiltrate into adipose tissues in a fat depot-specific manner. These results suggest that fat depot-specific differences in adipogenesis affect body fat distribution in fattening Wagyu.

Discipline: Animal Science
Additional key words: adipocyte, intramuscular, subcutaneous, visceral

Introduction

Adipose tissue growth induced by obesity (adipogenesis) occurs through adipocyte hyperplasia (increase in adipocyte number) and hypertrophy (increase in adipocyte size) (Prins & O'Rahilly 1997). Studies in humans and rodents have indicated fat depot-specific differences in adipogenesis (Fried et al. 2015). Fat depot-specific differences in adipogenesis are important factors in the production of beef cattle. The distribution of adipose tissues influences the carcass value of beef cattle. Subcutaneous and visceral fat content affect the carcass yield grades of beef cattle. The intramuscular fat content of longissimus muscle, called marbling, is crucial for determining the quality grades of beef. Japanese black (Wagyu) cattle are characterized by a greater ability to accumulate intramuscular adipose tissue than other cattle breeds, such as Angus and Holstein (Motoyama et al. 2016). In this review, we will discuss the fat depot-specific differences in the adipogenesis of fattening Wagyu.

Adipocyte size

The anatomical site of adipose tissue affects adipocyte size. Figure 1 shows adipocytes from subcutaneous, intramuscular, and visceral fat depots of fattening Wagyu cattle. The visceral adipocytes of Wagyu are larger than those of subcutaneous and intramuscular adipocytes. Adipose tissue accumulation is affected by adipocyte size. The marbling level of the

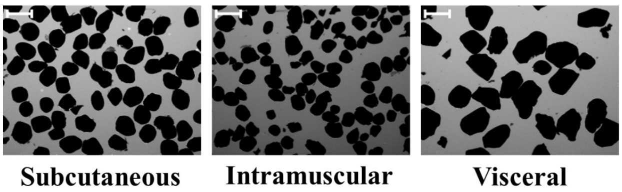

Fig. 1. Adipocytes from subcutaneous, intramuscular, and visceral depots of fattening Wagyu
The white scale bar indicates 300 µm (magnification: ×40).

*Corresponding author: toyamada@affrc.go.jp
Received 16 November 2023; accepted 26 February 2024.

longissimus muscle is represented by the beef marbling standard number (BMS No.) in Japan (Japan Meat Grading Association 2014). The BMS No. reflects the intramuscular adipose tissue content of the longissimus muscle (Iida et al. 2015, Yamada et al. 2020b). We showed that the BMS No. of fattening Wagyu was positively correlated with the size of the intramuscular adipocytes (Yamada et al. 2020b). These results indicate that the intramuscular fat accumulation is affected by adipocyte size. The breed of cattle also affects adipocyte size in a fat depot-specific manner. The body fat percentage of Wagyu was higher than that of Holstein (Yamada et al. 2009). We showed that the visceral adipocyte size of Wagyu was significantly larger than that of Holstein (Yamada et al. 2014). In addition, the intramuscular adipocyte size of Wagyu was significantly larger than that of Holstein (Yamada et al. 2022). These results indicate that breed differences between Wagyu and Holstein affect adipocyte size in visceral and intramuscular depots. In contrast, there was no difference in subcutaneous adipocyte size between Wagyu and Holstein (Yamada et al. 2014). These results indicate that breed differences between Wagyu and Holstein do not affect the adipocyte size in the subcutaneous depot.

Adipogenic transcription factors

Adipogenesis is regulated by the expression of adipogenic transcription factors (Fig. 2). Studies in humans and rodents indicated that CCAAT/ enhancer-binding protein (C/EBP) family (C/EBPα, C/ EBPβ, and C/EBPδ) and peroxisome proliferator-activated receptor γ (PPARγ) play essential roles during adipogenesis (Cristancho & Lazar 2011). C/EBPβ and C/ EBPδ are expressed in the early stage of adipogenesis. In contrast, C/EBPα and PPARγ are expressed in the late stage of adipogenesis. The anatomical site of adipose tissue is associated with the expression of adipogenic transcription factors. We studied the effects of fattening periods (at 19, 24, and 29 months of age) on the expression of adipogenic transcription factor protein in fattening Wagyu cattle (Yamada et al. 2007). The expressions of C/ EBPβ- liver-enriched activator protein in subcutaneous, intermuscular, and visceral adipose tissue at 29 months of age were significantly lower than those at 19 months. These results suggest that the preadipocyte differentiation capacity of subcutaneous, intermuscular, and visceral depots decrease as the fattening periods progress. In contrast, the expressions of C/EBPα protein in intermuscular adipose tissue at 29 months of age were significantly higher than those at 19 months. There were no differences in the C/EBPα expression level among fattening periods in subcutaneous and visceral depots. These results suggest that the fattening period affects the adipocyte terminal differentiation capacity in a fat depot-specific manner. The breed of cattle also affects the expression of adipogenic transcription factors. The expressions of C/EBPβ and C/EBPα proteins in the visceral adipose tissue of Wagyu were higher than those in Holstein (Yamada et al. 2009). In addition, the expressions of C/EBPβ and C/EBPα genes in the intramuscular adipose tissue of Wagyu were higher than those in Holstein (Yamada et al. 2022). These results suggest that breed differences in the adipogenic capacity of visceral and intramuscular depots between Wagyu and Holstein are affected by the expression of adipogenic transcription factors.

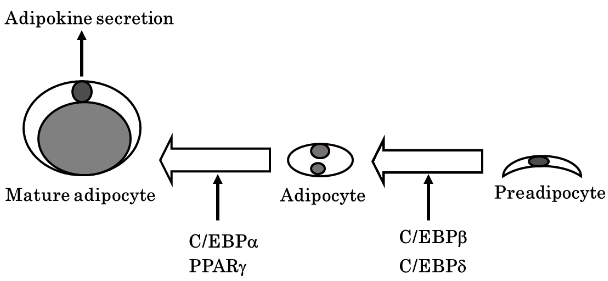

Fig. 2. Adipogenic transcription factor expression patterns during adipogenesis
C/EBPβ and C/EBPδ are expressed in the early stage of adipogenesis. C/EBPα and PPARγ are expressed in the late stage of adipogenesis.

Adipokines

Mature adipocytes secrete many hormones and cytokines, called adipokines (Fig. 2). Studies in humans and rodents indicate that adipokine secretions are affected by obesity (Luo & Liu 2016). We showed that the plasma leptin concentration positively correlates with body fat content in crossbred Wagyu steers (Yamada et al. 2003). The leptin mRNA expression level in adipose tissue positively correlates with adipocyte size in Wagyu (Yamada et al. 2010) and crossbred Wagyu steers (Yang et al. 2003). In addition, vascular endothelial growth factor (VEGF) and fibroblast growth factor-2 (FGF-2) mRNA expression levels in adipose tissues positively correlate with adipocyte size in Wagyu (Yamada et al. 2010). The anatomical site of adipose tissue affects adipokine gene expression. The expression level of leptin mRNA was higher in the visceral adipose tissue of Wagyu than in subcutaneous and intramuscular adipose tissues (Yamada et al. 2010). The expression levels of VEGF and FGF-2 mRNA in the visceral adipose tissue of Wagyu were also higher than those in subcutaneous and intramuscular adipose tissues (Yamada et al. 2010). These results suggest that fat depot-specific differences in adipokine gene expression are affected by adipocyte size.

Macrophage infiltration

In humans and rodents, obesity promotes the infiltration of macrophages into adipose tissues. Adipose tissue macrophages stimulate the chronic inflammation of adipose tissues (Russo & Lumeng 2018). The anatomical site of adipose tissue affects macrophage infiltration. We showed that macrophage infiltration into visceral adipose tissue was higher than that into subcutaneous and intramuscular adipose tissues in fattening Wagyu cattle (Yamada et al. 2018). The breed of cattle also affects macrophage infiltration into adipose tissues in a fat depot-specific manner. We showed that the macrophage infiltration into visceral adipose tissues of Wagyu was higher than that in Holstein. In contrast, there was no difference in the macrophage infiltration into subcutaneous adipose tissue between Wagyu and Holstein (Yamada et al. 2020a). These results indicate that breed differences between Wagyu and Holstein do not affect macrophage infiltration in subcutaneous depots. P53 is a master regulator of macrophage infiltration into adipose tissue in humans and rodents (Kung & Murphy 2016, Shimizu et al. 2012). We showed that the expression of the p53 gene in the visceral adipose tissue of Wagyu was higher than that of Holstein (Yamada et al. 2020a). We also showed no difference in the expression of the p53 gene in subcutaneous adipose tissue between Wagyu and Holstein (Yamada et al. 2020a). These results suggest that breed differences in macrophage infiltration into adipose tissue between Wagyu and Holsteins might be affected by p53 expression. Studies in humans and rodents show that macrophages stimulate adipose tissue fibrosis (Yao et al. 2022). In Wagyu cattle, *fat necrosis*, characterized by an increasing fibrous area within the visceral adipose tissue, occurs occasionally during fattening periods (Ito et al. 1968, Oka et al. 2015). These results suggest that higher macrophage infiltration into visceral depots might affect the adipose tissue fibrosis in fattening Wagyu.

Adipose tissue macrophages impaired the proliferation and differentiation of preadipocytes in humans and rodents (Liu et al. 2017, Nawaz et al. 2017). In addition, macrophage-induced adipose tissue fibrosis reduces adipogenic capacity in rodents (Tanaka et al. 2014). These results suggest that increased macrophage infiltration into visceral adipose tissue might trigger the limitation of adipogenic capacity in visceral depots of fattening Wagyu. Studies in rodents indicated that reducing the adipogenic capacity in white adipose tissue caused by macrophages induced a shift in the free fatty acid storage site from body fat to ectopic fat (Tanaka et al. 2014). Intramuscular adipose tissue is classified as ectopic fat. The intramuscular adipose tissue content of the longissimus muscle was positively correlated with the BMS No. in fattening Wagyu (Iida et al. 2015, Yamada et al. 2020b). We showed that the BMS No. of fattening Wagyu was negatively correlated with the visceral fat percentage (Yamada et al. 2020b). These results suggest that the fat storage site of fattening Wagyu with high intramuscular adipose tissue content shifts from visceral fat to intramuscular (ectopic) fat.

Conclusion

Figure 3 shows the proposed model of intramuscular fat accumulation in fattening Wagyu. Macrophage infiltration into visceral adipose tissue increases as the fattening periods progress. Macrophages in visceral fat stimulate the inflammation of adipose tissue and impair the adipogenic capacity of visceral depots. Reducing the fat storage capacity in the visceral depot induces the conversion of the free fatty acid storage site from the visceral to the intramuscular depot. As a result, intramuscular adipogenesis occurs through adipocyte hyperplasia and hypertrophy via the expression of adipogenic transcription factors. These results suggest

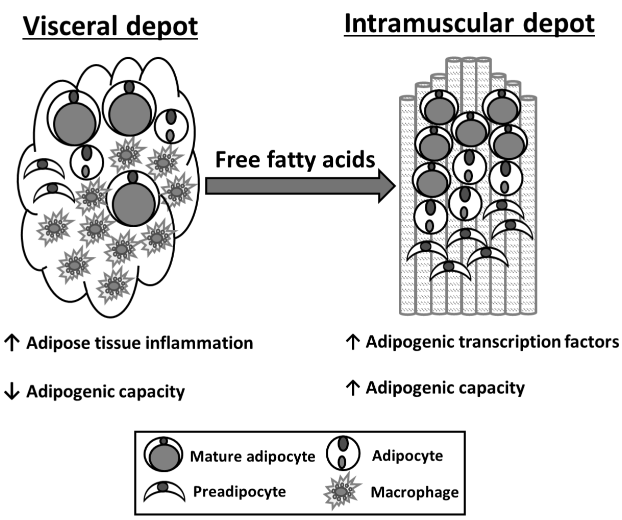

Fig. 3. Proposed model of intramuscular fat accumulation in fattening Wagyu
Macrophages in visceral fat stimulate the inflammation of adipose tissue and impair the adipogenic capacity of visceral depots. Reducing the adipogenic capacity in visceral depots induces the conversion of free fatty acid storage sites from visceral to intramuscular depots. Intramuscular adipogenesis occurs via the expression of adipogenic transcription factors.

that fat depot-specific differences in adipogenesis affect body fat distribution in fattening Wagyu.

References

Cristancho, A. G. & Lazar, M. A. (2011) Forming functional fat: a growing understanding of adipocyte differentiation. *Nat. Rev. Mol. Cell. Biol.*, **12**, 722-734.

Fried, S. K. et al. (2015) Shaping fat distribution: new insights into the molecular determinants of depot- and sex-dependent adipose biology. *Obesity*, **23**, 1345-1352.

Iida, F. et al. (2015) Effect of fat content on sensory characteristics of marbled beef from Japanese black steers. *Anim. Sci. J.*, **86**, 707-715.

Ito, T. et al. (1968) Pathological studies on fat necrosis (lipomatosis) in cattle. *Jpn. J. Vet. Sci.*, **30**, 141-150.

Japan Meat Grading Association (2014) Beef carcass trading standards. http://www.jmga.or.jp/standard/beef [In Japanese].

Kung, C. P. & Murphy, M. E. (2016) The role of the p53 tumor suppressor in metabolism and diabetes. *J. Endocrinol.*, **231**, R61-R75.

Liu, L. et al. (2017). Adipose tissue macrophages impair preadipocyte differentiation in humans. *PLoS One*, **12**, e0170728.

Luo, L. & Liu, M. (2016) Adipose tissue in control of metabolism. *J. Endocrinol.*, **231**, R77-R99.

Motoyama, M. et al. (2016) Wagyu and the factors contributing to its beef quality: a Japanese industry overview. *Meat Sci.*, **120**, 10-18.

Nawaz, A. et al. (2017). CD206^{+} M2-like macrophages regulate systemic glucose metabolism by inhibiting proliferation of adipocyte progenitors. *Nat. Commun.*, **8**, 286.

Oka, A. et al. (2015) Effects of clay on fat necrosis and carcass characteristics in Japanese black steers. *Anim. Sci. J.*, **86**, 878-883.

Prins, J. B. & O'Rahilly, S. (1997) Regulation of adipose cell number in man. *Clin. Sci.*, **92**, 3-11.

Russo, L. & Lumeng, C. N. (2018) Properties and functions of adipose tissue macrophages in obesity. *Immunology*, **155**, 407-417.

Shimizu, I. et al. (2012). p53-induced adipose tissue inflammation is critically involved in the development of insulin resistance in heart failure. *Cell Metab.*, **15**, 51-64.

Tanaka, M. et al. (2014). Macrophage-inducible C-type lectin underlies obesity-induced adipose tissue fibrosis. *Nat. Commun.*, **5**, 4982

Yamada, T. et al. (2003) The relationship between plasma leptin concentrations and the distribution of body fat in crossbred steers. *Anim. Sci. J.*, **74**, 95-100.

Yamada, T. et al. (2007) Effects of fattening periods on the expression of adipogenic transcription factors in Wagyu beef cattle. *Meat Sci.*, **76**, 289-294.

Yamada, T. et al. (2009) Expression of adipogenic transcription factors in adipose tissue of fattening Wagyu and Holstein steers. *Meat Sci.*, **81**, 86-92.

Yamada, T. et al. (2010) Fat depot-specific differences in angiogenic growth factor gene expression and its relation to adipocyte size in cattle. *J. Vet. Med. Sci.*, **72**, 991-997.

Yamada, T. et al. (2014) Fat depot-specific differences in pref-1 gene expression and adipocyte cellularity between Wagyu and Holstein cattle. *Biochem. Biophys. Res. Commun.*, **445**, 310-313.

Yamada, T. et al. (2018) Fat depot-specific differences of macrophage infiltration and cellular senescence in obese bovine adipose tissues. *J. Vet. Med. Sci.*, **80**, 1495-1503.

Yamada, T. et al. (2020a) Breed differences in macrophage infiltration and senescence state in adipose tissues of Wagyu and Holsteins. *Anim. Sci. J.*, **91**, e13443.

Yamada, T. et al. (2020b) Fat depot-specific effects of body fat distribution and adipocyte size on intramuscular fat accumulation in Wagyu cattle. *Anim. Sci. J.*, **91**, e13449.

Yamada, T. et al. (2022) Metabolomic analysis of plasma and intramuscular adipose tissue between Wagyu and Holstein cattle. *J. Vet. Med. Sci.*, **84**, 186-192.

Yang, S. H. et al. (2003) Fat depot-specific differences in leptin mRNA expression and its relation to adipocyte size in steers. *Anim. Sci. J.*, **74**, 17-21.

Yao, J. et al. (2022) Adipose tissue macrophage in obesity-associated metabolic diseases. *Front. Immunol.*, **13**, 977485.